Saskatchewan
Agriculture
and Food

ADF

AGRICULTURE DEVELOPMENT FUND

FINAL REPORT

20040493

DEVELOPMENT OF THE JAPANESE FEED MARKET FOR WESTERN CANADIAN BARLEY

Funded by: The Agriculture Development Fund

November 2008

Prepared by: University of Saskatchewan (U of S)

Development of Domestic and International Markets for Western Canadian Barley & Other Opportunity Feeds

Final Report

To

Saskatchewan Agriculture Development Fund

ADF Project # 20040493

By

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Milestone Date August 31, 2008

1.0 Executive Summary

The objective of this work is to develop a barley grain based feed product targeted to production of premium quality beef for domestic and international markets. This project was initiated by Japanese business interests (Takushoku Co., Ltd, and Rakuno Gakuen University) and our group (North East Terminals, Ltd., North West Terminals, Ltd., Pound-Maker Agventures, West Central Pelleting, and the University of Saskatchewan). Initial work by our group prior to this project, was to develop a pelleted barley grain/canola meal product. However, concerns relating to digestive upset (bloat and ruminal acidosis) resulting from rapid degradation of barley starch were evident with this initial product and improvements in processing methodology and feeding management information are required.

This work focused on feeding wheat-based DDGS in combination with barley to growing and finishing cattle. Wheat-based DDGS is potentially an ideal product for incorporation into a processed barley product. It is high in fibre and protein yet low in starch. Work with corn-based DDGS has shown it to be an excellent energy source for cattle, at least equal to corn grain. No work has been carried out with wheat-based DDGS for growing and finishing cattle.

Three trials were carried out to evaluate the effects of titrated levels of wheat-based dried distiller's grains with solubles (DDGS) on feedlot performance, carcass characteristics and rumen fermentation parameters of cattle. In trial 1, a barley grain-based diet (0% DDGS) was used as a control. It was formulated to 12% CP and 1.52 and 0.93 Mcal kg^{-1} net energy of maintenance (NE_m) and net energy of gain(NE_g) respectively, during the backgrounding period and to 13% crude protein (CP) and 1.90 and 1.26 Mcal kg^{-1} NE_m and NE_g respectively, during finishing. Wheat-based DDGS replaced on a dry matter basis (DM) barley grain at levels of 8, 16, 24 and 32% during backgrounding and 6, 12, 18 and 23% during finishing. During backgrounding dry matter intake ($P = 0.02$), ADG ($P = 0.04$), and ultrasound (US) *longissimus. dorsi* gain ($P = 0.02$) exhibited a cubic response to DDGS inclusion level with theoretical minima at 6.9, 8.1 and 6.9% DDGS respectively, and theoretical maxima responses at 27.2, 30.8 and 23.9% DDGS, respectively. Feed efficiency exhibited a quadratic response ($P = 0.02$) to DDGS inclusion level with a theoretical poorest response at 13.1% DDGS. Similar responses were noted during the first 56 d of the finishing period, however over the course of the finishing period no effect of DDGS inclusion level was noted on average daily gain (ADG), DMI, feed efficiency (FE), ultrasound measurements or on any carcass traits.

Trial 2 examined the effects of graded levels of wheat-based DDGS (0, 7, 14, 21% DM basis) on rumen fermentation characteristics using rumen cannulated heifers. Rumen pH measurements indicated that the pH mean at or below 5.8 and 5.5 decreased as DDGS inclusion level increased to 14% DM. The highest values ($P<0.05$) for pH area between the benchmarks of 5.5 and 5.2, pH area below 5.2 and time below pH 5.2 were found at the 14% DDGS inclusion level, pointing to rumen fermentation characteristics associated with severe rumen acidosis. Ammonia nitrogen levels, percent acetate, percent butyrate and the acetate: propionate ratio increased linearly ($P < 0.0001$) with DDGS. Propionate concentration decreased linearly ($P = 0.006$) as the level of DDGS increased.

In situ rumen degradation kinetics showed that the DM and CP soluble fraction of DDGS to be significantly higher than that of rolled barley. Similarly, effective degradability of dry matter (EDDM) and of crude protein (EDCP) were greater for barley. Trial 3 followed a similar experimental protocol and examined the effects of wheat-based DDGS in processed barley pellets on rumen fermentation parameters of rumen cannulated heifers. Rumen pH of cattle fed processed barley was lower than that of cattle fed rolled barley. Addition of 15 or 25% DDGS to a processed barley pellet did not improve rumen pH, particularly at pH cutoff points of 5.5 and 5.2, associated with mild to severe acidosis. Addition of 35% DDGs to a processed barley pellet improved rumen pH to levels associated with the rolled barley treatment. As in trial 2 rumen ammonia N levels increased with DDGS inclusion.

The results of this trial indicate that wheat-based DDGS has an energy value at least equal to that of barley grain with no adverse effects on cattle performance or carcass quality. Inclusion of wheat-based DDGS in a processed barley pellet, would generate a feed byproduct that would be at least equal to barley in energy content and would be significantly higher in protein. Based on the results of this trial, cattle feeders can replace up to 32% of rolled barley in backgrounding diets and 23% of rolled barley in finishing diets with wheat-based DDGS with no adverse affects on health or performance. The results of this work show however, that inclusion of a ground barley/DDGS pellet at levels typical of barley in finishing diets in North America would predispose cattle to sub-acute rumen acidosis. As such, the ideal market for such a product would be in backgrounding or growing rations where inclusion rates would be at lower levels and acidosis related problems would not be as great a concern.

2.0 Introduction:

The primary objective of this work is to develop a barley grain based feed product targeted to production of premium quality beef for domestic and international. This project was initiated by Japanese business interests (Takushoku Co., Ltd, and Rakuno Gakuen University) and our group (North East Terminals, Ltd., North West Terminals, Ltd., Pound-Maker Agventures, West Central Pelleting, and the University of Saskatchewan). Through our initial work, we have made progress with a barley grain/canola meal product. However, concerns relating to digestive upset (bloat and ruminal acidosis) resulting from rapid degradation of barley starch remain and improvements in barley processing methodology and feeding management information are required.

With respect to control of rumen acidosis and lower feed intakes which were the primary problems with our initial work in this area, the ideal feed to combine with barley for our export customers would be one that is high in energy and protein, low in fermentable carbohydrate and high in protein. Developing such a combination was one of the primary aspects of our research proposal. Recent developments in Saskatchewan's ethanol industry has provided us with an opportunity to evaluate such a feed in combination with barley. That feed is wheat-based dried distiller's grains with solubles (DDGS).

Husky Energy has expanded its ethanol production capacity. This includes a 130 million liter plant in Lloydminister, Sk. and a similar size plant in Minnedosa, Mb. It is well known that profitability of ethanol production is tied not only to production cost and market value of ethanol but also to the value obtained from byproduct sales (i.e. carbon dioxide, feed byproducts). Feed byproducts include wet (WDG) and dried distiller's grains with solubles (DDGS). The economic sustainability of incorporating ethanol production with cattle and the feeding of wet byproducts is well documented (i.e. Pound-Maker Agventures, Ltd., Lanigan, Sk). However, the size of the Husky Oil ethanol plants plus their competitive advantage with respect to drying costs, dictates that the majority of the distillers grains will be marketed as dried distillers grains with solubles (DDGS). This provides us however, with the opportunity to obtain a byproduct feed for inclusion in our processed barley pellet with the desired nutrient characteristics.

There has been extensive research on the feeding value of DDGS for livestock, with the majority applying to corn byproducts. This research (i.e. Boila and Ingals, University of Manitoba) has shown that dried distillers grains is an excellent protein supplement for dairy cows in early lactation and has been sold into this market at a premium. Feeding DDGS to beef cattle while nutritionally attractive, has not been economically justifiable to this point. The competitiveness of DDGS in beef cattle rations as well as for export markets is however, expected to change with the continued growth of the North American ethanol industry. It is logical to target beef cattle feeding enterprises both domestically and internationally as a potential market for this product. However, little research has been conducted with feeding DDGS to growing and finishing beef cattle, particularly with wheat-based products.

As indicated, at the University of Saskatchewan, we have a cooperative project with partners in western Canada and Japan to develop a value-added barley-based protein pellet that can be exported to Japan. The non-GMO status, high fibre, high energy and protein and low starch content of wheat-based DDGS would make it a very attractive protein supplement for this pellet if performance and cost of gain of cattle fed rations based on barley and wheat-based DDGS rations is equal or superior to that of conventional fed cattle.

There are several challenges that need to be met in order to develop a feed market for cattle for a barley / DDGS combination including:

- optimal level of barley / DDGS for growing & finishing cattle;
- composition and quality of DDGS;
- effects on carcass quality;
- pellet quality and processing characteristics
- rumen degradability characteristics of barley / distiller's grains and fermentation parameters.

As evident from the above discussion, there are numerous research questions that require answers, in order to fully develop a market for a value-added barley product in combination with DDGS and other byproducts available from secondary processing in Saskatchewan.

The hypothesis of our research is it is possible to develop domestic and international markets for western Canadian barley and byproducts of secondary processing including that from the ethanol industry as well as grain processing that focus on defined nutritional value and minimizing health problems such as acidosis.

The following work was undertaken to prove this hypothesis and provide our partners with data that they can use in the development of their feed business both domestically and internationally.

3.0 Trial 1: Effect of graded levels of wheat based dried distiller's grains on growth performance and carcass characteristics of feedlot steers.

The western Canadian ethanol industry has undergone substantial expansion in recent years. This sector of the industry differs from its counterparts in eastern Canada or the United States in that wheat instead of corn is the principle substrate used for fermentation, although blends of corn and wheat have been used depending on supply and price. The primary feed related byproduct derived from cereal grain-based ethanol production is dried distiller's grains with solubles (DDGS) and is the result of the complete drying of the whole stillage that remains after the ethanol has been distilled off (Larson et al. 1993). As such, DDGS are a concentrated source of CP, fibre, lipid and certain minerals such as phosphorous and sulfur (Larson et al. 1993; Boila and Ingalls, 1994a; Mustafa et al. 1999).

Performance information on cattle fed wheat-based DDGS is limited. Most of the research conducted to date has involved either wheat-based wet distiller's grains (Ojowi et al. 1997) or thin stillage (Fisher et al. 1999; Iwanchysko et al. 1999). These trials have shown that cattle fed these byproducts perform as well or better than traditional barley-fed cattle. With respect to wheat-based DDGS, Boila and Ingalls (1994a and 1994b) examined the rumen degradation characteristics of CP and post-ruminal availability of amino acids, however there has been no performance information published on growing and finishing cattle fed this byproduct. In contrast, there has been a considerable amount of research on the feeding value of corn-based DDGS. Benson et al. (2005) reported that corn-based DDGS can be included in feedlot finishing diets at levels up to 35 % of diet DM without negatively affecting performance, however carcass traits including subcutaneous (SC) fat thickness, weight and yield grade increased linearly as level of corn-based DDGS increased. Research has determined that corn-based wet distiller's grains plus thin stillage has a higher NE_g (2.53 Mcal kg^{-1}, Larson et al. 1993; 2.16 Mcal kg^{-1}, Ham et al. 1994) than both whole corn grain (1.55 Mcal kg^{-1}, NRC 1984) and corn-based DDGS (1.87 Mcal kg^{-1}, Ham et al. 1994), in part due to increased levels of fat in the by-product when compared to whole corn grain. Similar information is required on the relative energy value of wheat-based DDGS when replacing barley grain in growing and finishing diets.

In light of the increasing supply of wheat-based DDGS there is a need for research that targets the optimal level of this byproduct in the diet of growing and finishing cattle. The objectives of this study were to evaluate the performance and carcass quality characteristics of cattle fed graded levels of wheat-based DDGS and based on growth performance, determine the relative energy value of this by-product. Our hypothesis centered on the concept that replacing barley-grain in a ration balanced for crude protein with wheat-based DDGS would improve performance due to superior energy content of wheat-based DDGS.

3.1 MATERIALS AND METHODS

Animals Management and Feeding

Two hundred crossbred weaned steer calves (290 ± 17 kg) were purchased from a local auction market and shipped to the University of Saskatchewan Beef Cattle Research Unit. All calves were identified and processed on arrival including vaccination against clostridial disease, infectious bovine rhinotracheitis, and parainfluenza 3, implantation with Component E-S™ (Elanco, Guelph, ON) and treatment for parasites with Ivomec™ (MSD AgVet, Division of Merck Frosst Canada Inc., Kirkland, QC). Steers were stratified by weight and assigned to one of twenty outdoor pens. Each of the 20 pens was randomly assigned to one of five treatments. The trial consisted of an 85-d backgrounding period and a finishing period with an end-point of 625 kg live weight (unshrunk basis). Cattle were adapted to the finishing diet through a series of eight ration changes occurring every three days which involved a gradual increase in barley grain and DDGS in the diet through replacement of grass hay, barley straw and barley silage. Feed was delivered ad libitum, twice daily at 0800 and 1400. Cattle were cared for in accordance with the Canadian Council of Animal Care guidelines (CCAC, 1993).

Cattle were weighed on consecutive days at the start of test, a midpoint corresponding to the end of the backgrounding period and at the end of trial, to obtain average weights for start and end of backgrounding as well as the start and end of finishing. Upon the completion of the backgrounding period, the steers were re-implanted with Component TE-S™ (Elanco, Guelph, ON).

Cattle were fed ad libitum, twice daily. Daily feed consumption was measured on a pen basis. Every two weeks, feed bunks were cleaned and ort weight recorded. Samples of the total mixed rations were also collected every two weeks and frozen until analyzed. Grain and supplement samples were taken monthly and upon receipt of new loads. Forage samples were taken every two weeks for determination of DM content and stored for chemical analysis.

Ultrasound was used to measure live animal SC fat depth, and *l. dorsi* area, which were used to determine accretion rates. Animals were scanned monthly and immediately prior to slaughter using an Aloka 500 V real-time ultrasound machine equipped with a 17 cm linear array transducer according to Bergen et al. (1997).

Animals were slaughtered at XL Beef Inc. (Moose Jaw, SK). Carcass weight, yield and quality grades, SC fat thickness and *l. dorsi* area measurements were recorded by personnel of the Canadian Beef Grading Agency. Liver abscess scores were determined based on the Elanco scoring system as adapted by McKinnon et al. (1992).

Treatments and Dietary Composition

The backgrounding phase was designed to target a daily gain of 1.0 to 1.2 kg d^{-1}. During this period the control diet consisted of 45% concentrate (barley grain and supplement), and 55% forage (barley silage, brome grass hay and barley straw (DM basis; Table 1). For the wheat-based DDGS treatments DDGS replaced dry-rolled barley grain in the control diet at 8, 16, 24 and 32% (DM basis), respectively (Table 3.1). The control diet was formulated to 1.52 and 0.93 Mcal kg^{-1} of NE_m and NE_g, respectively. The finishing phase was designed to allow maximum gain through to the selected end-point. The control diet for the finishing phase consisted of 90% barley grain, 5% supplement and 5%

barley silage (DM basis) and was formulated to 1.90 and 1.26 Mcal kg^{-1} of NE_m and NE_g, respectively. For the wheat-based DDGS treatments, DDGS replaced barley grain at 6, 12, 18 and 23% (DM basis) (Table 3.2). Calcium to phosphorous ratios were formulated to range from 1.5:1 to 2:1 and all diets were formulated to meet or exceed NRC (1996) requirements and to contain 27 mg kg^{-1} of monensin sodium (Elanco Animal Health, Calgary, AB). Wheat-based DDGS was supplied in 3 separate loads by Husky Energy (Minnedosa, MB).

Chemical Analysis

Forage DM content was determined by oven drying samples at 55 °C for 48 h. Samples were then ground using a hammer mill with a 1-mm screen (Christie-Norris Laboratory Mill, Christie-Norris Ltd. Chelmsford, UK). For concentrate analysis, samples were ground using a Retsch ZM-1 grinder (Haan, Germany) with a 1-mm screen. Feed samples were analyzed for DM (AOAC method # 930.15) and CP (AOAC method # 984.13) according to (AOAC, 2000). Acid and neutral detergent fibre content was determined using an Ankom, 200 fibre analyzer (Ankom Technology, NY).

Each batch of wheat-based DDGS was analyzed according to AOAC (2000) by Cumberland Valley Analytical Services (CVAS, Hagerstown, MD). Samples were analyzed for DM (AOAC method # 930.15), CP (AOAC method # 990.03) with a Leco FP-528 Nitrogen Combustion Analyzer (St. Joseph, MI), ADF and ADIN (AOAC method # 973.18), ash (AOAC method # 942.05) and fat (AOAC method # 920.39). NDF content was analyzed using the method of Van Soest et al. (1991)

Statistical Analysis

The experiment was analyzed as a completely randomized design using the Proc Mixed Procedure of SAS 9.1.3 (SAS, Cary, NC) with pen as the experimental unit. The model included the fixed effect of treatment. Orthogonal polynomial contrasts were used to examine the linear, quadratic, cubic and quartic effects of DDGS inclusion rate. Significant responses ($P \leq 0.05$) were discussed using slope (linear) or theoretical minima / maxima (quadratic/cubic) of the best fit polynomial regression equation. Liver abscess scores and marbling data were analyzed using the GLIMMIX macro (SAS, Cary, NC) with a binomial error structure and logit data transformation.

3.2 RESULTS AND DISCUSSION

Our objective was to determine performance and carcass quality of cattle fed increasing levels of wheat-based DDGS and to test the hypothesis that replacing barley-grain with wheat-based DDGS in diets balanced to meet or exceed CP would improve performance due to superior energy content of the wheat-based DDGS. Analysis of the total mixed diets fed during backgrounding (Table 3.1) and finishing (Table 3.2) indicate CP levels for the control diet met or exceeded requirements for the targeted performance (NRC, 1996). As expected CP levels increased as DDGS inclusion rate increased. Similarly fibre levels (ADF & NDF) increased, reflecting the higher concentration of structural carbohydrates in DDGS relative to barley.

Crude protein content of the wheat-based DDGS averaged 37.2 ± 0.7% (DM basis) Table 3.3). This value is somewhat lower than that reported by Boila and Ingalls (1994a; average 40.7 ± 3.1%) for wheat-based DDGS but similar to that (35.7%) reported

by Thacker and Widyaratne (2007). Spiehs et al. (2002) reported 30.2 ± 6.4% CP for corn-based DDGS while Kleinschmit et al. (2007) reported an average CP value of 32.1%. Differences in CP content between wheat- and corn-based DDGS reflect differences in the nutrient content of the original cereal grain (Cromwell, 1993). This fact is also reflected in fibre and lipid content. In the current trial, NDF, ADF and ether extracted fat of wheat-based DDGS averaged 46.5 ± 1.2%, 13.2 ± 0.2% and 5.0 ± 0.3%, respectively. In contrast, Spiehs et al. (2002) reported average values for corn-based DDGS of 42.1 ± 14.3% for NDF, 16.2 ± 28.4% for ADF and 10.9 ± 7.8% for crude fat. Comparable NDF (32.9%) and ADF (16%) results were reported by Kleinschmit et al. (2007) for corn-based DDGS. The high fat value of corn-based DDGS is a contributing factor to its relatively high energy value for ruminants (Larson et al. 1993).

Acid detergent insoluble nitrogen (ADIN) has been found to be a good indicator of nitrogen digestibility in heat damaged forages (Nakamura et al. 1994). Yu and Thomas (1976) found that ADIN (% total nitrogen) was the best single indictor of total N digestion ($r^2 = 0.86$) out of 30 analytical determinations. Similar results were reported by Goering et al. (1972). The ADIN value for wheat-based DDGS in the current study averaged 10.5 ± 0.2% of total N. Boila and Ingalls (1994a) reported an average value of 11.8% ADIN for wheat-based DDGS. Kleinschmit et al. (2007) reported ADIN values for five corn-based DDGS samples to range from 7.5 to 23.1% of total N with an average of 12.4%. Factors such as amount of solubles added back, the extent of drying, and temperature affect the ADIN content and digestibility of DDGS (Van Soest, 1989; Weiss et al., 1989; Kleinschmit et al. 2007). It should be noted that while ADIN content has been used as an indicator of heat damage in forages, in non-forage protein sources such as DDGS, it has not been found to be a reliable indicator of protein digestibility (Nakamura et al. 1994).

Backgrounding Phase

During the backgrounding phase cattle fed the control diet gained an average of 1.2 kg d^{-1}, which met expectations based on formulated energy levels (NRC, 1996; Table 3.4). Animals fed the control diet had a DMI of 7.64 kg d^{-1} and a FE of 0.157. As DDGS inclusion level increased, a cubic effect ($P = 0.02$) on DMI was noted ($y = 7.62 - 0.102x + 0.009x^2 - 0.0002x^3$ $R^2 = 0.53$ SEP 0.27). Solving this equation for theoretical minima / maxima gives a minimum intake at a DDGS inclusion rate of 6.9% (DM basis) and a maximum intake at 27.2% DDGS. A similar response ($P = 0.04$) was noted for ADG ($y = 1.203 - 0.030x + 0.002x^2 - 0.00004x^3$ $R^2 = 0.55$ SEP 0.07) with a theoretical minima and maxima at 8.1 and 30.8% DDGS (DM basis), respectively. Feed efficiency exhibited a quadratic ($P = 0.02$) effect ($y = 0.157 - 0.0008x + 0.000032x^2$ $R^2 = 0.37$ SEP 0.006) with poorest efficiency at 13.1% DDGS (DM basis). Previous research has noted positive responses for ADG, DMI and FE in cattle backgrounded with various levels of corn-based DDGS (Ham et al. 1994; Klopfenstein, 1996) and wheat-based WDG (Ojowi et al. 1997). In the present study, it is not clear why DMI and ADG were reduced at the low inclusion rates of DDGS yet improved at higher levels. It is possible that dietary energy values change as DDGS inclusion levels change. Stock et al. (2000) in summarizing data from several independent trials showed that while the energy value of corn-DDGS averaged 109% of corn grain, actual energy value (83 to 124%) depended on dietary inclusion rate and generally peaked as dietary inclusion levels increased. Interactions

between dietary ingredients, particularly that of fermentable carbohydrate, rumen degradable protein and fat content could influence overall ration digestibility which could give the appearance that DDGS are higher in energy content at moderate inclusion levels than at low or high levels. The results of this study indicate that further work should be undertaken with backgrounding cattle to determine if these results are repeatable or simply an inherent result of the current trial.

Ultrasound measurements taken at the end of the backgrounding period showed no ($P > 0.05$) effect of treatment on US fat measurements, however US *l. dorsi* gain exhibited a cubic response ($P = 0.02$) to increasing DDGS inclusion level ($y = 0.152 - 0.0082x + 0.0008x^2 - 0.00002x^3$ $R^2 = 0.31$ SEP 0.03) (Table 3.4) with theoretical minimum and maximum responses at 6.9 and 23.9% DDGS (DM basis), respectively. The goal of backgrounding is to minimize fat accretion and promote both frame and muscle development. As evidenced from Table 3.4, the SC fat and *l. dorsi* area accretion rates indicate that this goal was achieved in the present study.

Finishing Period

Finishing period ADG, DMI, and FE averaged 1.85 ± 0.04 kg d^{-1}, 11.02 ± 0.14 kg d^{-1}, and 0.168 ± 0.02, respectively and was not ($P > 0.05$) influenced by treatment (Table 3.5). These results are in contrast to corn-based DDGS studies which have shown improvements in finishing DMI and ADG (Benson et al. 2005) as well as FE (Larson et al. 1993) as DDGS inclusion rate increased. With respect to the corn-based research, it has been shown that corn-based DDGS has a higher energy value relative to corn grain (1.87 Mcal kg^{-1} vs. 1.55 Mcal kg^{-1} NEg), accounting for the superior performance of cattle fed DDGS (Ham et al. 1994). In the current study, finishing performance was not influenced (positively or negatively) when wheat-based DDGS replaced barley grain in the finishing diets at levels up to 23 % of the diet DM. These results would indicate that at the levels used in the finishing phase of this study, wheat-based DDGS had similar NE_m and NE_g values to barley grain (i.e. 2.00 to 2.06 Mcal kg^{-1} NE_m & 1.34 to 1.40 Mcal kg^{-1} NE_g; NRC, 1996). Lack of treatment effects ($P > 0.05$) on ultrasound SC fat and *l. dorsi* area measurements at the end of the finishing period support this conclusion (Table 3.5).

Carcass Traits

Liver abscess scores were not ($P > 0.05$) affected by treatment (Table 3.6). Similar results for finishing cattle fed corn-based DDGS were reported by Ham et al. (1994). It is somewhat surprising to not see an effect of DDGS inclusion level on liver abscess scores, as substituting DDGS for barley grain should in theory reduce the possibility of SARA and associated problems such as liver abscesses (Larson et al. 1993; Ham et al. 1994). There was no effect ($P > 0.05$) of treatment on hot carcass weight or on measured carcass traits including dressing percentage, marbling score, SC fat thickness, *l. dorsi* area or yield grade (Table 6). These results are similar to Ojowi et al. (1997) and Buckner et al. (2007) who found no negative or positive influence of feeding wet wheat- or corn-based distiller's grains, respectively, on carcass characteristics of finishing cattle. In contrast, Benson et al (2005) noted that cattle fed corn-based DDGS produced carcasses with more SC fat and poorer yield grades. There have also been some carcass quality issues associated with feeding either wet or dry corn-based distiller's grains at high inclusion

rates (50 % of diet DM), in particular with regard to reduced shelf life of the meat (Roeber et al. 2005). This may be a result of the high levels of unsaturated fat associated with corn-based DDGS and has not been reported as a problem with beef cattle fed wheat-based WDG (Shand et al. 1998).

3.6 CONCLUSION

Supplementing graded levels of wheat-based DDGS to backgrounding cattle resulted in minimum responses in both DMI and ADG at theoretical DDGS inclusion levels of 6.9 and 8.1% (DM basis) respectively and theoretical maximum responses at 27.2 and 30.8% DDGS (DM basis), respectively. Feed efficiency exhibited a quadratic response to DDGS inclusion level with theoretical poorest conversions at 13.1% DDGS (DM basis). The relatively poor performance during backgrounding at low inclusion levels and superior performance at higher levels indicates that wheat-based DDGS may interact with other dietary ingredients to alter dietary energy content depending upon the level fed. Further research is required to define the response of backgrounding cattle to this byproduct under varying feeding regimes. No effect of increasing DDGS inclusion level was noted on finishing performance or on carcass quality. The results of this trial indicate that wheat-based DDGS can be used to replace barley in both backgrounding and finishing diets supplying both energy and protein and other nutrients such as phosphorus and sulphur without negatively impacting performance and that for finishing cattle, the energy value of wheat-based DDGS is at least equal to that of barley grain.

3.7 ACKNOWLEDGEMENTS

Appreciation is expressed to the staff of the University of Saskatchewan Beef Cattle Research Unit for the care of the cattle used in these studies. The authors would like to thank the Agriculture Development Fund of the Saskatchewan Ministry of Agriculture for funding this project and to Husky Energy Ltd. of Calgary, AB, for supplying the wheat-based DDGS used in these trials. Appreciation is also expressed to Drs. H. Block and P. Yu for assistance with statistical analysis.

Table 3.1 Diet composition and analysis for growing cattle fed graded levels of wheat-based DDGS

	DDGS Inclusion Level (% DM)				
	0	8	16	24	32
Diet Composition (% DM basis)					
Barley silage	24.6	24.5	24.4	24.3	24.2
Brome hay	14.0	13.9	13.8	13.7	13.7
Barley straw	15.0	15.0	15.0	14.9	14.8
Supplement	7.7	7.6	7.6	7.6	7.5
Barley grain	38.7	30.8	23.0	15.2	7.6
Wheat DDGS	0.0	8.2	16.2	24.3	32.2
Supplement Composition (% DM basis)					
Barley grain	0	56.2	56.2	56.2	56.2
Canola Meal	66.7	-	-	-	-
Limestone	11.1	21.9	21.9	21.9	21.9
Vitamin premix[Z]	8.8	8.7	8.7	8.7	8.7
Trace mineral salt[Y]	5.0	4.9	4.9	4.9	4.9
Rumensin Premix[X]	4.9	4.8	4.8	4.8	4.8
Tallow	3.5	3.5	3.5	3.5	3.5
Ration Analysis (% DM basis)[W]					
CP	12.6 ± 0.5	14.5 ± 0.6	16.5 ± 0.4	18.4 ± 0.3	20.3 ± 0.2
ADF	27.1 ± 0.6	28.1 ± 0.4	29.0 ± 0.3	30.0 ± 0.3	30.9 ± 0.4
NDF	38.4 ± 0.5	39.8 ± 0.6	41.2 ± 0.8	42.6 ± 1.0	44.0 ± 1.3

[Z] 445,000 IU vitamin A, and 88,000 IU vitamin D_3 kg^{-1}.
[Y] Rumensin premix: 3% monensin sodium.
[X] Trace mineral salt: 95% NaCl, 12 000 ppm Zn, 10 000 ppm Mn, 4 000 ppm Cu, 400 ppm I, 60 ppm Co, 30 ppm added Se.
[W] Values shown with standard error of means.

Table 3.2 Diet composition and analysis for finishing cattle fed graded levels of wheat-based DDGS

	DDGS Inclusion Level (% DM)				
	0	6	12	18	23
Diet Composition (% DM basis)					
Barley silage	5.3	5.2	5.2	5.2	5.2
Supplement	5.5	5.8	5.7	5.7	5.7
Barley grain	89.2	83.1	77.4	71.6	65.8
Wheat DDGS	0.0	5.9	11.7	17.5	23.3
Supplement Composition (% DM basis)					
Barley grain	11.8	45.8	45.8	40.8	40.8
Tallow	3.5	3.5	3.5	3.4	3.4
Canola Meal	38.3	-	-	-	-
Limestone	21.9	26.2	26.2	31.4	31.4
LS106[Z]	9.6	9.6	9.6	9.6	9.6
Rumensin Premix[Y]	7.2	7.2	7.2	7.2	7.2
Trace mineral salt[X]	7.7	7.7	7.7	7.6	7.6
Ration Analysis (% DM basis)[W]					
CP	13.2 ± 1.4	14.5 ± 1.3	15.9 ± 1.2	17.3 ± 1.2	18.7 ± 1.1
ADF	12.8 ± 0.7	13.8 ± 0.7	14.7 ± 0.6	15.7 ± 0.6	16.6 ± 0.6
NDF	17.7 ± 0.8	18.7 ± 0.8	19.7 ± 0.7	20.6 ± 0.7	21.6 ± 0.6

[Z] 445,000 IU vitamin A, and 88,000 IU vitamin D_3 kg^{-1}.
[Y] Rumensin premix: 3% monensin sodium.
[X] Trace mineral salt: 95% NaCl, 12 000 ppm Zn, 10 000 ppm Mn, 4 000 ppm Cu, 400 ppm I, 60 ppm Co, 30 ppm added Se.
[W] Values shown with standard error of means.

Table 3.3 Nutrient composition of wheat-based DDGS used in the Growing and Finishing Trials (DM basis)

	Batch 1	Batch 2	Batch 3	SE[z]
DM	93.4	93.9	94.3	0.26
CP	37.2	38.5	36.0	0.72
NDF	45.1	48.9	45.6	1.19
ADF	13.5	13.3	12.9	0.18
ADIN[Z]	10.8	10.7	10.1	0.21
Ash	4.7	4.1	4.2	0.18
Ether Extract	4.5	4.8	5.6	0.33

[z]SE = standard error.

Table 3.4 Effects of feeding graded levels of wheat-based DDGS on the performance of backgrounding cattle

	DDGS Inclusion Level (% DM)						*P* value	*P value* Contrast			
	0	8	16	24	32	PSEM[z]	Treatment	Linear	Quadratic	Cubic	Quartic
Body Weight (kg)											
Start of Backgrounding	290.7	290.4	289.5	290.2	290.5	0.26	-	-	-	-	-
End of Backgrounding	393.0	383.1	388.1	397.2	399.3	3.01	0.01	0.02	0.02	0.04	0.96
Average Daily Gain (kg)	1.20	1.09	1.16	1.26	1.28	0.04	0.01	0.01	0.04	0.04	0.92
Dry Matter Intake (kg)	7.64	7.26	7.71	7.97	7.94	0.14	0.01	0.01	0.31	0.02	0.45
Gain : Feed	0.158	0.150	0.151	0.158	0.161	0.003	0.05	0.12	0.02	0.22	0.64
Ultrasound SC Fat (mm)											
Start of Test	1.80	1.98	1.48	1.85	1.60	0.22	-	-	-	-	-
End of Backgrounding	2.93	2.63	2.45	3.18	2.68	0.24	0.27	0.95	0.52	0.09	0.18
Gain ($mm\ d^{-1}$)	0.01	0.01	0.01	0.02	0.01	0.003	0.35	0.46	0.54	0.08	0.90
Ultrasound l. dorsi area (cm^2)											
Start of Test	57.21	59.04	57.27	56.63	58.38	0.75	-	-	-	-	-
End of Backgrounding	70.31	69.02	71.24	70.00	69.61	1.01	0.62	0.90	0.67	0.41	0.20
Gain ($cm^2\ d^{-1}$)	0.15	0.12	0.16	0.16	0.13	0.01	0.07	0.92	0.51	0.02	0.11

[z]PSEM = Pooled standard error of the mean

Table 3.5 Effects of feeding graded levels of wheat-based DDGS on the performance of finishing cattle

	DDGS Inclusion Level (% DM)					*P value*		*P value* Contrast			
	0	6	12	18	23	PSEM[z]	Treatment	Linear	Quadratic	Cubic	Quartic
Body Weight (kg)	608.5	606.1	601.1	607.2	612.4	2.81	0.13	0.69	0.32	0.14	0.04
Average Daily Gain (kg)	1.87	1.86	1.77	1.88	1.88	0.04	0.34	0.86	0.16	0.83	0.12
Dry Mater Intake (kg)	11.02	10.90	10.85	11.25	11.09	0.14	0.33	0.29	0.48	0.18	0.26
Gain : Feed	0.170	0.171	0.164	0.167	0.169	0.002	0.21	0.47	0.11	0.33	0.14
Ultrasound Subcutaneous Fat (mm)											
End of Finishing	8.70	8.58	8.25	9.00	8.70	0.47	0.85	0.81	0.69	0.59	0.39
Gain (mm d^{-1})	0.05	0.05	0.05	0.05	0.05	0.003	0.67	0.33	0.30	0.77	0.72
Ultrasound l. dorsi area (cm^2)											
End of Test	98.19	98.87	98.84	97.97	98.17	1.35	0.98	0.83	0.73	0.68	0.86
Gain (cm^2 d^{-1})	0.24	0.25	0.23	0.24	0.26	0.02	0.54	0.50	0.26	0.46	0.37

[z]PSEM = Pooled standard error of the mean.

Table 3.6 Effects of feeding graded levels of wheat-based DDGS on carcass characteristics of finishing cattle

	DDGS Inclusion Level (% DM)						*P value*	*P value* Contrast			
	0	6	12	18	23	PSEM[z]	Treatment	Linear	Quadratic	Cubic	Quartic
Hot Carcass Weight (kg)	361.4	365.1	361.6	362.6	361.0	1.92	0.62	0.64	0.39	0.46	0.32
Subcutaneous fat (mm)	8.6	8.6	9.4	9.3	9.1	0.55	0.68	0.70	0.74	0.17	0.80
Longissimus dorsi area (cm^2)	99.5	101.8	99.2	99.2	98.9	1.89	0.82	0.50	0.71	0.45	0.58
Carcass : Live weight Ratio	58.9	59.6	59.6	58.7	58.9	0.33	0.19	0.40	0.12	0.11	0.48
Yield Grade (%)	61.0	61.1	61.5	61.0	61.4	0.55	0.89	0.43	0.65	0.69	0.80
Marbling Score (% of Cattle)											
AAA (Small or >)	18.0	22.5	27.5	27.5	10.0	7.3	0.47	-	-	-	-
AA (Slight)	71.8	75.0	67.5	70.0	87.5	8.2	0.52	-	-	-	-
A (Traces)	7.7	0.0	2.5	2.5	0.0	2.0	1.00	-	-	-	-
B2 (Dark)	2.5	2.5	2.5	0	2.5	2.0	1.00	-	-	-	-
Liver Abscess Sccore[y]											
0 (% of cattle)	71.0	76.5	83.3	58.1	65.4	9.7	0.43	-	-	-	-
1 (% of cattle)	12.9	14.7	5.6	12.9	0.0	3.6	0.55	-	-	-	-
2 (% of cattle)	12.9	5.9	5.6	12.9	19.2	6.2	0.55	-	-	-	-
3 (% 0f cattle)	3.2	2.9	5.6	16.1	15.4	5.9	0.41	-	-	-	-

[z]PSEM = Pooled standard error of mean.

[y] Liver abscess scoring: 0 = no abscess; 1 = one small abscess; 2 = two to four small to medium (<2.54 mm) abscesses; 3 = one or more large (>2.54 mm) abscesses or greater than four small to medium abscesses.

4.0 Trial 2: Effect of graded levels of wheat-based DDGS on rumen fermentation and rumen degradation kinetics in finishing cattle.

Acidosis is a major concern in high performance ruminants. It is caused by the consumption of large amounts of ruminally-degradable carbohydrate, low amounts of effective fibre, or both (Noeck, 1997). Numerous papers have described the problem of acidosis (Dunlop and Hammond 1965; Dunlop 1972; Slyter 1976) however; the more economically important problem is chronic or sub-acute acidosis (SARA; Dirksen 1970; Nocek 1997; Owens 1998; Penner *et al.* 2007). Cattle with SARA may not appear sick, but their feed intake and therefore their performance is reduced (Owens, 1998). Reduced performance due to erratic feeding behavior and SARA is believed to cost as much as $15 to $20 per animal in lost efficiency (Schwartzkopf-Genswein *et al.* 2003). Acidosis related problems can further affect the profitability of feeding cattle due to the increased incidence of liver condemnations, reduced weight gains, decreased feed efficiency and decreased carcass yield (Nagaraja and Chengappa, 1998).

Although SARA involves a lowering of rumen pH below pH 5.8 (Maekawa *et al.* 2002; Penner *et al.* 2007), it is not adequate to simply define it as being caused by low pH. SARA is primarily determined by the physical characteristics of the diet, specifically the proportion of fibre that is of adequate particle length to maintain proper rumen function (Yang *et al.* 2001). By ensuring adequate amounts of physically effective fibre (minimum particle size of 1.18mm; Mertens, 1997), rumination is stimulated which results in greater time spent chewing and increased buffering capacity due to the secretion of saliva to the rumen. Therefore, it is often assumed that time spent chewing is a good indication of rumen health (Yang *et al.* 2001).

In western Canadian feedlots, barley (*Hordeum vulgare*) is the common cereal grain used in growing and finishing diets due to abundant supply and relative low cost (Boss and Bowman, 1996). Processing barley results in increased rumen starch digestibility, increased average daily gain, decreased feed to gain ratio and decreased acetate to propionate ratio (Bevans *et al.* 2005). However, there are drawbacks to feeding grain which has been excessively processed. These include an increase in the rate of starch fermentation which predisposes cattle to metabolic diseases including bloat, acidosis, laminitis and liver abscesses (Owens *et al.* 1998; Yang *et al.* 2001). In such situations, feedstuffs which invoke minimal rumen upset, maintain or increase feed intake, and lead to decreased days to market would be an appealing option (Stock *et al.* 1990).

The development of the ethanol industry in Canada has resulted in a growing supply of ethanol by-products. Ethanol by-products such as wet distiller's grains (WDG) and dry distiller's grains with solubles (DDGS) contain high protein, high fibre (NDF) and low starch content (Ham *et al.* 1994). Previous research has shown that cattle fed corn-based distiller's by-products showed increased ADG and improved feed efficiency. It was hypothesized that the increased feed efficiency was in part a result of a reduction in the incidence of sub-acute rumen acidosis (Firkins *et al.* 1985; Larson *et al.* 1993; Ham *et al.* 1994). There has been no research in this regard concerning wheat-based DDGS. Considering that barley is a highly digestible feed relative to corn (Beauchemin *et al.* 1994), replacing barley with wheat-based DDGS may be an attractive feeding strategy from the view point of preventing SARA.

The objectives of this study were to characterize the effects of graded levels of wheat-based DDGS on rumen pH, fermentation characteristics and eating behavior as indicators of rumen health. A second objective was to characterize the *in vitro* rumen degradation kinetics of wheat-based DDGS.

4.1 MATERIALS AND METHODS

Animals, Housing and Experimental Design

Four spayed heifers (388 ± 25kg) were surgically fitted with soft plastic, 10 cm ruminal cannulas (Bar Diamond, Parma, ID). Cattle were housed in individual 3 x 3 m floor pens with rubber matting and individual water bowls. Each animal was randomly assigned to one of four dietary treatments in a 4 X 4 Latin square design. Each 28 d period included a 14 d dietary adaptation period, followed by a 6 d voluntary intake period (d 15 – 20) and a collection period during which chewing behavior was observed for 24 h (d 21), rumen contents where sampled over a 24 h period (d 23), and *in-dwelling* pH data collected over three consecutive 23 h periods (d 26 – 28).

Following the rumen metabolism study, two of the heifers (538 ± 38 kg) were maintained under the same conditions for an *in situ* digestibility trial. *In situ* nylon bag incubations of rolled barley and three different wheat-based DDGS samples were carried out over a 72 h period. All cattle were cared for in accordance with the Canadian Council of Animal Care guidelines (CCAC 1993).

Treatments and Dietary Composition

Prior to the start of the trial all four heifers were fed a barley based finishing ration (90 % concentrate, 5 % forage, 5 % supplement; DM basis). During the adaptation phase of each period, dietary transition occurred every three days by replacing 25 % of the barley or wheat-based DDGS until the final dietary treatment composition was reached. Cattle were fed daily at 0800 and 1600. Each morning, feed bunks were cleaned and the remaining feed weighed and recorded daily.

The control diet (0 % DDGS) consisted of 89 % barley grain, 6 % supplement and 5% barley silage (DM basis). Treatments included 7, 14 and 21 % DDGS replacing barley in the ration (DM basis; Table 4.1). All diets were formulated to meet NRC (1996) requirements for minerals and vitamins and to contain 28 mg $kg^{-1}$of monensin sodium (DM basis; Elanco Animal Health, Calgary, Alberta, Canada). Samples of dietary ingredients were taken for each period and frozen at -20 ° C for further chemical analysis.

Rumen Collections

Twenty-four h rumen fluid collections were started at 0800 on d 23 of each period with samples collected every 2 h. A representative rumen fluid sample was collected by sampling 500 mL of rumen fluid from the reticulum, ventral and caudal sacs of the rumen, as well as the rumen mat. These samples were then combined and strained through four layers of cheese cloth to remove particulate matter. Immediately after straining, duplicate measurements of pH were taken with a Model 265A portable pH meter (Orion Research Inc., Beverly, MA). From the collected fluid, three 5 mL aliquots of rumen fluid were then sub-sampled. One was preserved for VFA analysis by adding 1mL of 25 % (wt vol^{-1}) HPO_3, another for ammonia concentration by adding 1 mL H_2SO_4

and the final sub-sample was taken for osmolality analysis and stored without the addition of a preservative. All samples were stored at -20 °C in sealed plastic vials until analysis.

Volatile Fatty Acid Analysis

Samples stored for VFA analysis were first thawed and then centrifuged at 14000 rpm for 15 min in a Microfuge® 18 Microcentrifuge (Beckman Coulter™, Palo Alto, CA). The supernatant was then pipetted into 12 x 75 mm tubes and 1 mL crotonic acid (1 mg mL^{-1}) was added as an internal standard. Samples were filtered using a 0.45 µm filter and glass syringe and placed into two vials for duplicate analysis. Acetate, propionate, isobutyrate, butyrate, isovalerate, and valerate were identified and quantified in each sample using an Agilent 6890 Series GC System including an Agilent 7683 Series injector fitted with an Agilent Technologies High Performance GC Capillary Column (30.0 m x 320 µm x 0.25 µm, all Wilmington, DE). Total VFA concentration was determined by summing the concentrations of all individual acids (Ghorbani *et al.* 2002; Beauchemin *et al.* 2003a). A standard curve was prepared using standards purchased from Nu-Chek Prep, Inc. (Elysian, MN) and used to calculate the molar proportion of each of the previously mentioned acids.

Rumen Ammonia and Osmolality

Ruminal ammonia was determined using the phenol-hypochlorite method (Broderick and Kang 1980). Osmolality was determined using a Vapro™ Vapor Pressure Osmometer (Model 5520; Wescor Inc., Logan, Utah). Non-acidified samples were first centrifuged at 2000 rpm for 15 min using a Beckman Centrifuge (Model TJ-6; Palo Alto, CA). Each sample was analyzed in duplicate.

In-dwelling pH Measurement

In-dwelling pH collections were carried out from d 26 through d 28 of each period using the Indwelling Continuous pH Measurement System (Dascor, Escondido, CA) as described by Penner *et al.* (2006). Data loggers and pH probes were weighted and positioned within the ventral sac of the rumen to record pH over a 23 h period at 30 s intervals. Probes were removed daily between 0700 and 0800, cleaned, standardized (pH 4 and 7), and the pH data downloaded for analysis. Ruminal pH data over the 23 h was averaged for each minute and summarized as minimum pH, mean pH, and maximum pH. Animals were characterized as being in a state of ruminal acidosis (RA) as per the following pH profiles: mild (pH 5.8 - 5.5); moderate (pH 5.5 - 5.2); and acute (pH < 5.2) (Nocek 1997; Penner *et al.* 2007). In addition, the duration (min d^{-1}) and total area (pH*min) that pH was below each threshold was calculated.

Feeding Behaviour

Feeding behavior was recorded starting at 0800 on d 21 of each period. Animal behavior was recorded over a 24 h period at 5 minute intervals under one of the following categories: eating, ruminating, drinking, lying, or standing according to Yang *et al.* (2000). It was assumed that the behavior observed over the 5 min intervals lasted the entire time between observations. Observation methods were based on those of Beauchemin *et al.* (2001) and Maekawa *et al.* (2002a and 2002b).

Chemical Analysis

Forage DM content was determined by oven drying samples at 55 ºC for 48 h. Samples were then ground using a hammer mill with a 1 mm screen (Christie-Norris Laboratory Mill, Christie-Norris Ltd. Chelmsford, UK). For chemical analysis, concentrate samples of original undigested grain and DDGS treatments and pooled rumen *in situ* residues were ground using an ultra centrifugal mill (Retsch ZM 100, Haan, Germany) with a 1 mm screen. Dry matter, CP (Kjehldahl), and ash analysis was in accordance with AOAC (1990). Neutral detergent fiber and ADF analysis were carried out with an Ankom 200 fibre analyzer™ (Ankom Technology, NY). Particle size was determined using the Penn State particle separator and the procedure of Heinrichs and Kononoff (2002). The pH of DDGS and barley were determined using the procedure of Jasaitis (1987).

Statistical Analysis

Rumen fermentation parameters (pH, VFA concentration, osmolality, ammonia) and ingestive behavior were analyzed using the PROC MIXED procedure of SAS 9.1 (SAS Institute Inc. 2003), accounting for repeated measures with the random effect of heifer and the fixed effect of treatment. Polynomial orthogonal contrasts were used to determine significant linear, quadratic and cubic effects. The Kenward Roger adjustment on denominator degrees of freedom was used and significance declared at $P < 0.05$. Trends are discussed for $P < 0.10$. Regression analysis was carried out on significant contrasts to determine equations for linear, quadratic and cubic effects of DDGS inclusion rates. Furthermore equations were used to determine theoretical minima and maxima for each significant parameter.

4.2 RESULTS AND DISCUSSION

The design of this study tested the hypothesis that addition of wheat-based DDGS to barley based finishing diets would minimize acidic conditions in the rumen due to its high fibre and low starch nature. Wheat-based DDGS was supplied for this trial by the Husky Energy facility in Minnedosa, Manitoba. Details on the chemical composition of the wheat-based DDGS were given in (Beliveau and McKinnon, submitted). The control diet consisted of 95 % barley-based concentrate and 5 % barley silage (DM basis; Table 4.1) and was formulated to 12.0 % CP, and 1.52 and 0.93 Mcal kg^{-1} of NE_m and NE_g (DM basis). In treatments two through four, wheat-based DDGS was substituted for barley at 7, 14, and 21 % (DM basis). Formulated CP levels for the treatment diets were 13.2, 14.9, and 16.6 % (DM basis), respectively. Dry matter intake recorded over d 14 to 21 of each period averaged 11.2 ± 0.82 kg d^{-1} with no effect ($P = 0.51$) of treatment (Table 4.2).

Rumen pH (spot sample and in-dwelling)

Rumen pH measurements were taken with an in-dwelling pH probe as well with the traditional spot sampling method (Kononoff *et al.* 2003; Penner *et al.* 2006). Figure 4.1 shows that rumen pH measured over the course of the day, for all treatments behaved in a typical diurnal pH pattern, falling after feeding and then recovering (Penner *et al.* 2006). Mean ruminal pH averaged over the course of a 24 h period was not different between treatments regardless of the method of measurement used (Table 4.2). Failure to see a treatment effect was unexpected as the substitution of a non-starch concentrate for a

starch-based concentrate should theoretically result in reduced microbial fermentation activity, altered VFA production pattern including lower overall levels and increased rumen pH (Ørskov, 1986).

Rustomo (2006) suggested that mean ruminal pH or a single pH value at any one point in time is not a good indication of the extent or severity of SARA. Several authors have proposed that the length of time over a 24 h period that rumen pH is below a critical value is more important. Critical pH values proposed include time below 5.8 (mild), 5.6 (moderate) and below 5.2 (severe; acute; Nocek, 1997; Krause *et al.* 2002; Penner *et al.* 2007). Table 4.2 gives the results of the data analyzed in this fashion. Mean rumen pH at the cutoff of pH 5.8 ($P < 0.02$) and 5.5 ($P < 0.003$) decreased in a cubic fashion as DDGS inclusion level increased to 14 % DM, and then increased at the highest DDGS level. Regression analysis for pH 5.8 ($y = 5.40 + 0.048x - 0.011x^2 + 0.0004x^3$; $R^2 = 0.46$ SEP = 0.19) and pH 5.5 ($y = 5.23 + 0.043x - 0.0096x^2 + 0.0004x^3$; $R^2 = 0.52$ SEP = 0.15) shows that for these parameters the theoretical highest rumen pH is found at DDGS inclusion level of 2.5 and 2.6 % (DM) respectively. Theoretical minimum rumen pH was found to be at 14.8 % DDGS inclusion for pH 5.8 and 14.7 % (DM) DDGS for pH 5.5. A similar response ($P = 0.02$) was noted for the pH area under the curve between 5.5 and 5.2 ($y = 6895.4 - 1523.2x + 305.78x^2 - 11.364x^3$ $R^2 = 0.49$ SEP = 4296.8) with a theoretical minima and maxima at 3.0 and 15.0 % DDGS (DM basis), respectively. Time spent at or below pH 5.2 ($P = 0.04$) showed a cubic response ($P = 0.03$) with the greatest area under the curve at the 14 % (DM) DDGS and then dropping off at the 21 % inclusion level ($y = 259.04 - 78.887x + 16.486x^2 - 0.6175x^3$ $R^2 = 0.52$ SEP = 226.3). Solving this equation for theoretical minima / maxima gives minimum time below pH 5.2 at a DDGS inclusion rate of 2.9 % (DM basis) and maximum time below pH 5.2 at 15.0 % DDGS. These results, particularly the time below pH 5.2, are indicative of rumen conditions associated with acute acidosis in cattle fed the two intermediate DDGS diets (Penner *et al.* 2007).

Regression analysis indicated that rumen pH levels for each critical pH value (5.8, 5.5 and 5.2) were predicted to be highest and therefore optimal for rumen fermentation at DDGS inclusion levels of 2.5 to 3.0 %. Whereas the most acidic rumen pH was found to be at 14.7 – 15.0 % (DM) DDGS inclusion. Both of the intermediate treatments in this experiment were in the range between the theoretical minima and maxima for all in-situ pH parameters measured in this experiment. The addition of wheat-based DDGS at levels between 3.0 and 15.0 % resulted in a progressively more acidic rumen and therefore would negatively impact rumen fermentation through disruption of the microbial populations.

Fermentation Characteristics (VFA, Ammonia and Osmolality)

Figure 4.2 indicates that the diurnal pattern of total VFA concentration was highest ($P < 0.0001$) 2 to 4 hours post-feeding for each treatment and then declined in an inverse pattern to rumen pH (Figure 4.1). This pattern is consistent with feeding barley-grain based diets and has been shown by other researchers (Krause *et al.* 1998). Percent acetate ($P = 0.02$; $y = 51.218 + 0.3036x$ $R^2 = 0.13$ SEP = 5.4) and butyrate ($P = 0.04$; $y = 8.2262 + 0.1439x$ $R^2 = 0.07$ SEP = 3.5) increased in a linear fashion as DDGS inclusion level increased. In contrast propionate ($P = 0.01$; $y = 40.909 - 0.4964x$ $R^2 = 0.18$ SEP = 7.1) decreased in a linear fashion with increasing levels of DDGS. As a result, acetate to propionate ratio increased in a linear fashion ($P = 0.01$; $y = 1.2111 + 0.0496x$ $R^2 = 0.17$

SEP = 0.68) as DDGS inclusion level increased (Table 3.4). These changes in the molar proportion of VFA's are typical of diets with increasing forage content and likely reflect the relative high fibre nature of wheat-based DDGS relative to barley grain (Pitt *et al.* 1996; Russell, 1998).

Rumen osmolality levels were not affected by treatment (Table 4.3) and daily variation mirrored the pattern of total VFA over the course of the 24 h sampling period (Table 4.2). Ruminal NH_3 N values averaged 5.8 mg dL^{-1} which is within the range reported by Kang-Meznarich and Broderick (1980) for optimal fermentation (3.3 to 8.5 mg dL^{-1}). Rumen ammonia levels increased (P = 0.02) in a linear fashion as DDGS inclusion level increased (y = 4.8236 + 0.0914x R^2 = 0.07 SEP = 2.6). This result is a reflection of the higher CP content of wheat-based DDGS relative to barley and the fact that dietary CP levels increased from 11.8 to 16.4 % as DDGS inclusion level increased. It should be noted that even though rumen NH_3 N levels increased in a linear fashion, the magnitude of the increase relative to the control diet was not large (4.8 vs. 6.8 mg dL^{-1}) particularly in relation to differences in the CP content of the diets. This is likely a reflection of the fact that the RUDCP content of wheat-based DDGS is significantly higher than that of barley grain (Boila and Ingalls, 1994; Mustafa *et al.* 2000).

Feeding Behaviour

There was no difference in the time spent ruminating however there was a linear increase (y = 97.25 + 2.1964x R^2 = 0.30 SEP = 27.5) in time spent eating (P = 0.03) as DDGS inclusion level increased (Table 4.4). Total time spend drinking (average 13.5 ± 3.5 min), lying (average 927.3 ± 22.1 min) and standing (average 311 ± 15.1 min) were similar between treatments. Several researchers have suggested that the total time spent chewing (eating + ruminating) is positively correlated with saliva secretion and rumen buffering capacity (Beauchemin *et al.* 1994; Maekewa *et al.* 2002a and 2002b; Yang and Beauchemin 2006). The lack of a treatment effect suggests that the fibre content of DDGS is not effective at stimulating chewing activity (eating + ruminating). Therefore the fibre is ineffective at stimulating salvia production and does little to enhance rumen buffering capacity. This supports the finding of the rumen metabolism trial in that minimal rumen buffering is consistent with the increased time below pH 5.5 and 5.2 with increased DDGS in the diet (Mertens, 1997).

From the point of view of the hypothesis of this study, the results of the rumen metabolism trial were unexpected. Ham *et al.* (1994) postulated that substitution of corn-based DDGS for corn grain resulted in a reduction in SARA and consequently led to superior ADG and feed efficiency. In this study, substitution of wheat-based DDGS at 7 and 14 % of diet DM actually led to rumen pH conditions that are reflective of acute acidosis, while substitution at 21 % of the diet DM led to similar rumen pH values as that of the control fed animals. Our hypothesis was that since DDGS is a low starch and high fibre product, the opposite would happen (i.e. rumen pH would be maintained or increase as DDGS levels increased).

The severity of acidosis observed in this trial may be explained by the characteristics of DDGS that affect the buffering capacity of the rumen including those that affect saliva flow, extent and rate of rumen fermentation and feed particle passage rate. Particle separation analysis with the Penn State Particle Separator, using the method of Heinrichs and Kononoff (2002), showed that 74 ± 5 % of the wheat-based DDGS was

less than 1.18 mm in size. The remainder of the DDGS was retained on the 1.18 mm screen. The NDF content of the wheat-based DDGS used in this trial was 46.5 ± 2.1% (Beliveau and McKinnon, submitted). Mertens (1997) suggested that physically effective fibre which stimulates chewing and rumen buffering can be determined by multiplying the proportion of particles retained on a 1.18 mm sieve against the NDF content of the material sieved. Mertens (1997) suggested that the PE fibre needs to be at least 22 % of the diet to be effective in a dairy ration. Using this procedure the peNDF content of this by-product is ~ 11 %. Thus, although the wheat-based DDGS is a high fibre feed, it is unable to contribute to chewing activity and as a result, rumen buffering. This also explains why rumen pH was not increased at increased DDGS inclusion levels, and why feeding behavior was not different between treatments. Fibre of adequate particle size (>5mm) has been shown to promote chewing and rumination in dairy herds and thereby maintains rumen health through buffers secreted in salvia and moderated pH (Clark and Armentano, 2002; Yang and Beauchemin, 2006). Since the DDGS used in this trial is virtually all less than 8 mm in length, the fibre content of the wheat-based DDGS can be classified as non-physically effective fibre. Furthermore, a reduction in forage particle size has been reported to reduce the total time spent chewing and increase the rumen particulate passage rate (Welch, 1982; Kononoff and Heinrichs, 2003). Therefore, despite high fibre and low starch qualities of wheat-based DDGS it was ineffective in buffering rumen acid production in this trial.

The fact that rumen pH was negatively affected by DDGS inclusion, particularly the time spent below pH 5.2 for 7 and 14 % inclusion levels may also reflect the chemical nature of the by-product. Several workers have reported that the pH of cereal grain derived by-products from ethanol production is low (Giger-Reverdin *et al.* 2002). Jasaitis *et al.* (1987) reported that the initial pH of corn-based DDGS was 4.35 while that of barley grain was 5.73. In this study, the initial pH of wheat-based DDGS was 4.31 while that of barley grain was 5.36. Jasaitis *et al.* (1987) reported that the titratable acid (i.e. the amount of HCl required to drop pH to 4.0) for corn-based DDGS was 8 (meq x 10^{-3}). This compared to 60 for barley and 240 for alfalfa hay. This indicates that feeds such as DDGS and barley grain are poor rumen buffers at low pH levels. The fact that wheat-based DDGS has such a low initial pH may help explain why no benefit is seen in terms of rumen pH when DDGS is added to the diet. This may also help explain the cubic response to DDGS inclusion levels for time below pH 5.2. Adding DDGS to the diet increased the time below this critical pH associated with acute acidosis through the 7 and 14 % inclusion levels, which is consistent with its acidic nature and poor buffering ability.

This does not however explain the decrease in the time below pH 5.2 with the 21 % DDGS addition rate. While this observation is difficult to explain, it may be that at this level of DDGS, starch has been sufficiently replaced with a non-starch, high fibre feed that rumen fermentation is modified to the extent where pH is similar to the control diet. This conclusion is supported by changes in the molar proportions of VFA which showed that acetate increased in a linear fashion while propionate levels decreased with increased DDGS inclusion and A:P ratio widened. These findings are characteristic of rumen fermentation conditions associated with higher fibre diets (Russell, 1998).

Based on the results of this experiment, DDGS does not appear to be a good substitute for barley grain from the perspective of improving rumen fermentation

conditions indicative of SARA. Further research needs to be conducted to determine if acidosis mitigation is increased as the level of DDGS increases in the diet beyond 21 % of diet DM.

4.3 CONCLUSION

The results of this study show that supplementation of wheat-based DDGS with barley grain does not lead to rumen fermentation conditions that minimize problems with SARA. Despite the supplementation of wheat-based DDGS for barley, all treatments were equally susceptible to SARA. There was no difference between treatments when comparing overall mean pH. However as DDGS approached an inclusion level of 14 % (DM basis), rumen fermentation conditions became more acidic and indicative of SARA. This was indicated by mean pH below 5.8, 5.5, longest time spent below pH 5.2. As DDGS inclusion levels increased pH to 21 % rumen pH conditions improved to control levels, likely reflecting the high fibre, low starch nature of this by-product. Acetate, butyrate and A:P showed a linear increase while propionate decreased as DDGS levels increased indicating a change in the rumen fermentation away from starch digestion and towards fibre fermentation. Ammonia nitrogen levels increased linearly with the increase of nitrogen supplied in the diet by DDGS. These shifts can likely be attributed to the removal of highly digestible barley starch and higher levels of dietary fibre and N seen as wheat-based DDGS increased in the diet.

The results of this experiment indicate that ability of DDGS's to mitigate conditions indicative of SARA is less than that of barley grain. Analysis of the nutrient characteristics of wheat-based DDGS showed that it is an acidic feedstuff with a very small particle size. The natural acidity combined with a lack of treatment effect on rumen buffering activities such as chewing and ruminating resulted in a decrease in rumen pH with the addition of DDGS to the diet. At the highest levels of DDGS inclusion however the metabolic parameters such as molar proportions of VFA's and time spent below pH thresholds showed a trend towards improved rumen buffering capacity. Further research is necessary to determine the causation of the pH and VFA changes seen at 23 % DDGS inclusion level and to what inclusion level does this trend continue.

4.4 ACKNOWLEDGEMENTS

Appreciation is expressed to the staff of the University of Saskatchewan Livestock Research Unit for the care of the cattle used in these studies. The authors would like to thank the Agriculture Development Fund of the Saskatchewan Ministry of Agriculture for funding this project and to Husky Energy Ltd. of Calgary, AB, for supplying the wheat-based DDGS used in these trials.

Table 4.1 Diet composition and analysis for cattle fed titrated levels of wheat-based DDGS

	DDGS Inclusion Level (% DM)			
	0 %	7 %	14 %	21 %
Diet Composition (% DM basis)				
Silage	5.4	5.4	5.4	5.3
Supplement	5.7	5.6	5.6	5.6
Barley	88.9	81.8	74.7	67.8
Wheat DDGS	0.0	7.2	14.3	21.2
Supplement Composition (% DM basis)				
Barley	12.4	45.8	45.8	40.8
Tallow	0.0	3.5	3.5	3.4
Canola Meal	39.1	0.0	0.0	0.0
Limestone	22.9	26.2	26.2	31.5
LS106[Z]	10.1	9.6	9.6	9.6
Rumensin Premix[Y]	7.6	7.2	7.2	7.2
Trace mineral salt[X]	8.0	7.7	7.7	7.6
Ration Analysis (% DM basis)[W]				
CP	11.8 ± 1.3	13.0 ± 1.1	14.7 ± 1.1	16.4 ± 1.4
ADF	9.2 ± 1.1	9.52 ± 1.0	9.8 ± 0.8	10.1 ± 0.8
NDF	18.5 ± 2.2	20.5 ± 1.8	22.4 ± 1.7	24.4 ± 1.9

[Z] LS 106 44 500 IU vitamin A, and 88 000 IU vitamin D_3 kg^{-1}.

[Y] Rumensin premix: 3% monensin sodium.

[X] Trace mineral salt: 95% sodium chloride, 12 000 ppm zinc, 10 000 ppm manganese, 4 000 ppm copper, 400 ppm iodine, 60 ppm cobalt, 30 ppm added selenium.

[W] Values shown with standard error of means.

Table 4.2 Dry matter intake and rumen pH measurements of cattle fed titrated levels of wheat-based DDGS.

	DDGS Inclusion Level						*P* – Value Contrasts*		
	0 %	7 %	14 %	21 %	SEM	$P_{Treatment}$	*Linear*	*Quadratic*	*Cubic*
DMI (kg)	11.97	10.65	11.37	10.80	0.76	0.44	0.34	0.55	0.26
Mean Daily Rumen pH									
In-dwelling pH	5.81	5.79	5.50	5.84	0.14	0.21	0.68	0.19	0.12
Spot Sample pH	5.90	5.78	5.66	5.85	0.14	0.32	0.48	0.13	0.45
Rumen pH Parameter 5.8 or lower									
Mean pH	5.54	5.37	4.92	5.43	0.08	0.02	0.07	0.08	0.02
Total time (min)	764.8	725.7	923.3	696.9	119.9	0.45	0.99	0.24	0.20
Time between 5.5 and 5.8 (min)	172.8	227.4	137.6	303.5	56.77	0.25	0.26	0.08	0.15
pH Area between 5.5 and 5.8 (pH*sec)	9992	11349	16875	8669.2	2672	0.18	0.89	0.20	0.14
Rumen pH Parameter 5.5 or lower									
Mean pH	5.34	5.22	4.86	5.29	0.05	0.003	0.02	0.01	0.004
Time between 5.2 and 5.5 (min)	276.6	218.4	168.4	199.1	56.53	0.70	0.37	0.84	0.81
pH Area between 5.2 and 5.5 (pH*sec)	3914	7693	17443	3999	1931	0.02	0.24	0.05	0.02
Rumen pH Parameter 5.2 or lower									
Mean pH	5.10	4.88	4.89	5.05	0.07	0.21	0.72	0.72	0.82
Time below 5.2 (min)	259.0	302.9	691.4	153.9	112.9	0.04	0.87	0.05	0.03
pH Area below 5.2 (pH*sec)	2085	6882	14126	2085	3153	0.05	0.68	0.19	0.11

* Contrast analysis considering linear (L), quadratic (Q), and cubic (C) for treatment.

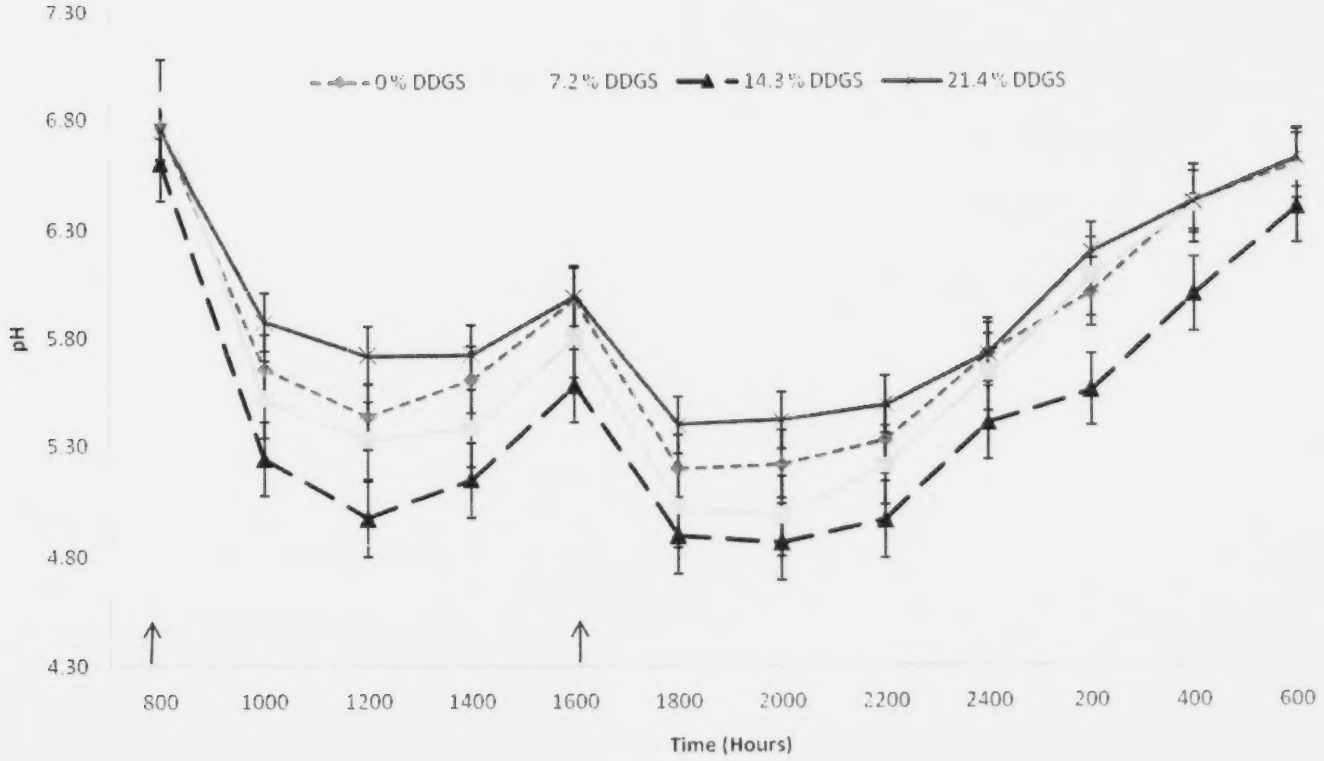

Figure 4.1 Effects of titrated levels of wheat-based DDGS on mean pH as measured with continuous in-dwelling monitors. Arrows represent feeding times.

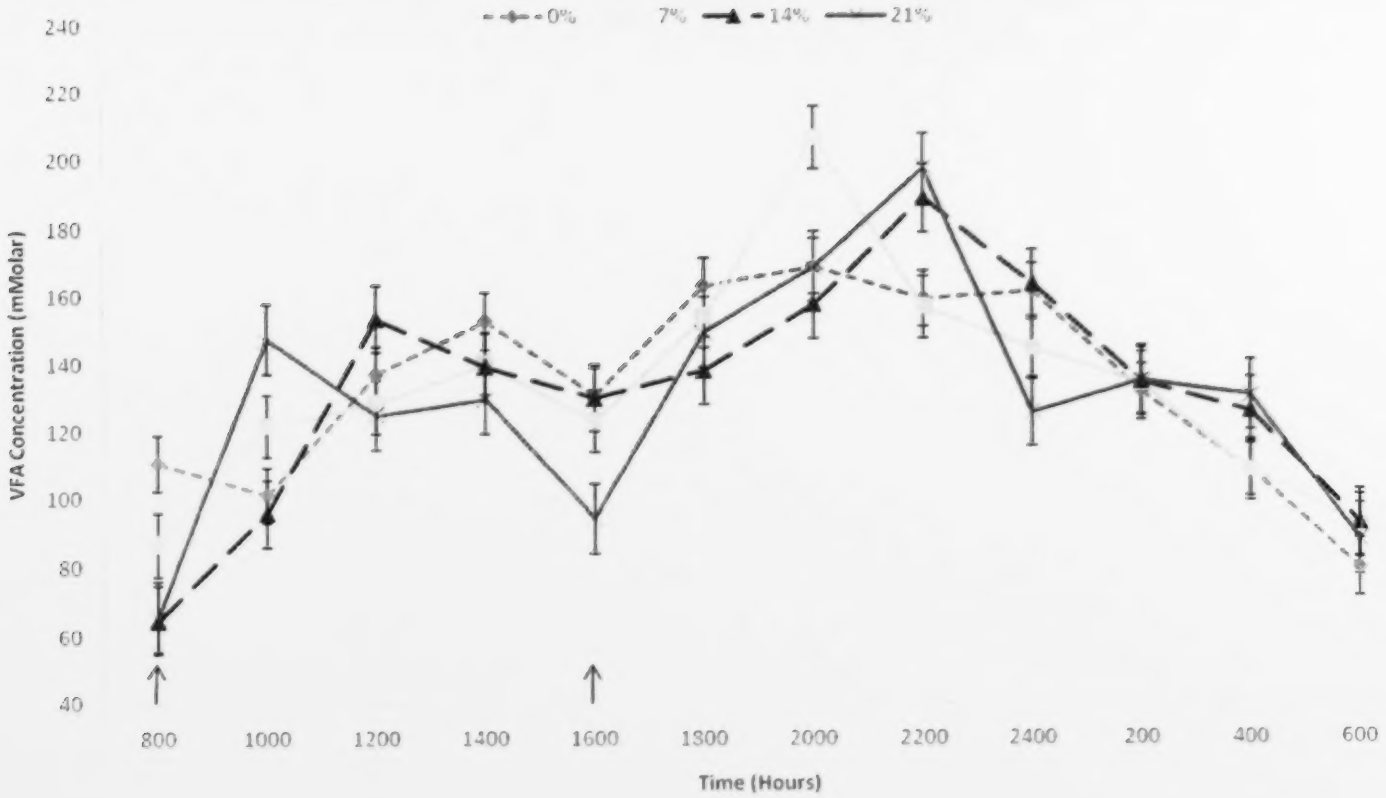

Figure 4.2 Effects of titrated levels of wheat-based DDGS on rumen total VFA concentrations. Arrows represent feeding times.

Table 4.3 Effects of feeding titrated levels of wheat-based DDGS on rumen fluid characteristics.

	DDGS Inclusion Level							*P* – Value Contrasts*		
	0 %	7 %	14 %	21 %	SEM	$P_{Treatment}$	P_{Time}	Linear	Quadratic	Cubic
Total VFA	136.0	132.5	131.2	129.5	8.95	0.96	<0.0001	0.62	0.93	0.95
Acetate (%)	51.4	52.7	54.6	56.6	1.54	0.14	0.056	0.02	0.84	0.94
Propionate (%)	40.8	38.8	34.4	32.3	2.04	0.04	0.04	0.006	0.99	0.61
A:P Ratio	1.3	1.4	1.7	2.1	0.19	0.03	0.04	0.01	0.44	0.99
Butyrate	8.2	8.9	10.1	10.9	0.88	0.20	<0.0001	0.04	0.96	0.81
IsoButyrate	0.2	0.2	0.2	0.2	0.02	0.72	0.02	0.68	0.32	0.74
Valerate	1.7	1.6	1.8	1.9	0.27	0.89	<0.0001	0.50	0.77	0.88
IsoValerate	0.4	0.5	1.5	0.6	0.29	0.13	0.41	0.23	0.12	0.07
Osmolality (mOsm L^{-1})	299.4	295.7	289.5	300.1	7.73	0.76	<0.0001	0.91	0.38	0.59
Ammonia (mg dL^{-1})	4.8	5.4	6.1	6.8	0.44	0.02	<0.0006	0.003	0.89	0.91

* Contrast analysis considering linear (L), quadratic (Q), and cubic (C) for treatment significance ($P < 0.05$) and trends ($P < 0.15$).

Table 4.4 Effect of titrated levels of wheat-based DDGS on the ingestive behavior for heifers fed a finishing ration.

	Wheat-based DDGS Inclusion Levels (% DM)					*P*-value	Regression Analysis		
	0 %	7 %	14 %	21 %	PSEM	Treatment	Linear	Quadratic	Cubic
Time (min day^{-1})									
Eating	111	93	126	151	13.46	0.07	0.03	0.14	0.34
Ruminating	368	294	331	280	43.79	0.52	0.28	0.80	0.33
Chewing[z]	479	386	476	431	45.09	0.53	0.73	0.48	0.23
Drinking	20	13	14	19	6.18	0.79	0.93	0.34	0.86
Lying	958	926	925	902	38.42	0.79	0.36	0.91	0.77
Standing	288	323	314	318	42.63	0.94	0.68	0.72	0.77

[z] Chewing = eating + ruminating

5.0 Trial 3: Effect of Pelleted Barley and Wheat-based DDGS on Rumen Fermentation Parameters

Initial studies in this project have shown that wheat-based DDGS has a similar energy value to barley grain for growing and finishing cattle. Inclusion of wheat-based DDGS in backgrounding (up to 32% DM basis) and finishing rations (up to 23% DM basis) showed that feedlot performance and carcass quality were not adversely affected. However, rumen fermentation studies indicated that at levels up to 21% of ration DM, DDGS did not improve rumen fermentation conditions associated high barley-based finishing diets.

Processing barley by grinding and pelleting is important to the development of export markets such as Japan for Canadian feed products. However it is known that highly processed barley is very rapidly digested in the rumen and further puts strain on the ability of the rumen to regulate pH (McKinnon and Williams unpublished). If development of a pelleted barley / wheat-based DDGS is to be successful, it is necessary to more fully characterize the effects of wheat-based DDGS on rumen fermentation parameters, particularly when fed in graded levels with ground and pelleted barley.

The objectives of this study were to characterize the effects of ground and pelleted barley in combination with graded levels of wheat-based DDGS on rumen pH and fermentation characteristics as indicators of rumen health.

5.1 MATERIALS AND METHODS

Animals, Housing and Experimental Design

Five spayed heifers (388 ± 25kg) were surgically fitted with soft plastic, 10 cm ruminal cannulas (Bar Diamond, Parma, ID). Cattle were housed in individual 3 x 3 m floor pens with rubber matting and individual water bowls. Each animal was randomly assigned to one of five dietary treatments in a 5 X 5 Latin square design. Each 28 d period included a 14 d dietary adaptation period, followed by a 6 d voluntary intake period (d 15 – 20) and a collection period during which rumen contents where sampled over a 24 h period (d 23), and *in-dwelling* pH data collected over three consecutive 23 h periods (d 26 – 28). All cattle were cared for in accordance with the Canadian Council of Animal Care guidelines (CCAC 1993).

Treatments and Dietary Composition

Prior to the start of the trial all five heifers were fed a barley based finishing ration (90 % concentrate, 5 % forage, 5 % supplement; DM basis). Cattle were fed daily at 0800 and 1600. Each morning, feed bunks were cleaned and the remaining feed weighed and recorded daily. The control diet (0 % DDGS) consisted of 88.5% rolled barley grain (RB). 5.5% supplement and 6% barley silage (DM basis). Treatments included the control ration with pelleted barley (PB) substituted for rolled barley or pelleted barley combined with 15 (PB15%DDGS), 25 (PB25%DDGS) or 35 % (PB35%DDGS) wheat-based DDGS (DM basis; Table 5.1). Pelleting was carried out at the University of

Saskatchewan feed mill with barley ground through a 6.25 mm screen prior to pelleting. All diets were formulated to meet NRC (1996) requirements for minerals and vitamins and to contain 28 mg $kg^{-1}$of monensin sodium (DM basis; Elanco Animal Health, Calgary, Alberta, Canada). Samples of dietary ingredients were taken for each period and frozen at -20 ° C for further chemical analysis.

Rumen Collections

Twenty-four h rumen fluid collections were started at 0800 on d 23 of each period with samples collected every 2 h. A representative rumen fluid sample was collected by sampling 500 mL of rumen fluid from the reticulum, ventral and caudal sacs of the rumen, as well as the rumen mat. These samples were then combined and strained through four layers of cheese cloth to remove particulate matter. Immediately after straining, duplicate measurements of pH were taken with a Model 265A portable pH meter (Orion Research Inc., Beverly, MA). From the collected fluid, three 5 mL aliquots of rumen fluid were then sub-sampled. One was preserved for VFA analysis by adding 1mL of 25 % (wt vol^{-1}) HPO_3, another for ammonia concentration by adding 1 mL H_2SO_4 and the final sub-sample was taken for osmolality analysis and stored without the addition of a preservative. All samples were stored at -20 °C in sealed plastic vials until analysis.

Volatile Fatty Acid Analysis

Samples stored for VFA analysis were first thawed and then centrifuged at 14000 rpm for 15 min in a Microfuge® 18 Microcentrifuge (Beckman Coulter™, Palo Alto, CA). The supernatant was then pipetted into 12 x 75 mm tubes and 1 mL crotonic acid (1 mg mL^{-1}) was added as an internal standard. Samples were filtered using a 0.45 µm filter and glass syringe and placed into two vials for duplicate analysis. Acetate, propionate, isobutyrate, butyrate, isovalerate, and valerate were identified and quantified in each sample using an Agilent 6890 Series GC System including an Agilent 7683 Series injector fitted with an Agilent Technologies High Performance GC Capillary Column (30.0 m x 320 µm x 0.25 µm, all Wilmington, DE). Total VFA concentration was determined by summing the concentrations of all individual acids (Ghorbani *et al.* 2002; Beauchemin *et al.* 2003a). A standard curve was prepared using standards purchased from Nu-Chek Prep, Inc. (Elysian, MN) and used to calculate the molar proportion of each of the previously mentioned acids.

Rumen Ammonia and Osmolality

Ruminal ammonia was determined using the phenol-hypochlorite method (Broderick and Kang 1980). Osmolality was determined using a Vapro™ Vapor Pressure Osmometer (Model 5520; Wescor Inc., Logan, Utah). Non-acidified samples were first centrifuged at 2000 rpm for 15 min using a Beckman Centrifuge (Model TJ-6; Palo Alto, CA). Each sample was analyzed in duplicate.

In-dwelling pH Measurement

In-dwelling pH collections were carried out from d 26 through d 28 of each period using the Indwelling Continuous pH Measurement System (Dascor, Escondido, CA) as

described by Penner *et al.* (2006). Data loggers and pH probes were weighted and positioned within the ventral sac of the rumen to record pH over a 23 h period at 30 s intervals. Probes were removed daily between 0700 and 0800, cleaned, standardized (pH 4 and 7), and the pH data downloaded for analysis. Ruminal pH data over the 23 h was averaged for each minute and summarized as minimum pH, mean pH, and maximum pH. Animals were characterized as being in a state of ruminal acidosis (RA) as per the following pH profiles: mild (pH 5.8 - 5.5); moderate (pH 5.5 - 5.2); and acute (pH < 5.2) (Nocek 1997; Penner *et al.* 2007). In addition, the duration (min d^{-1}) and total area (pH*min) that pH was below each threshold was calculated.

Feeding Behaviour

Feeding behavior was recorded starting at 0800 on d 21 of each period. Animal behavior was recorded over a 24 h period at 5 minute intervals under one of the following categories: eating, ruminating, drinking, lying, or standing according to Yang *et al.* (2000). It was assumed that the behavior observed over the 5 min intervals lasted the entire time between observations. Observation methods were based on those of Beauchemin *et al.* (2001) and Maekawa *et al.* (2002a and 2002b).

Chemical Analysis

Forage DM content was determined by oven drying samples at 55 ºC for 48 h. Samples were then ground using a hammer mill with a 1 mm screen (Christie-Norris Laboratory Mill, Christie-Norris Ltd. Chelmsford, UK). For chemical analysis, concentrate samples of original undigested grain and DDGS treatments and pooled rumen *in situ* residues were ground using an ultra centrifugal mill (Retsch ZM 100, Haan, Germany) with a 1 mm screen. Dry matter, CP (Kjehldahl), and ash analysis was in accordance with AOAC (1990). Neutral detergent fiber and ADF analysis were carried out with an Ankom 200 fibre analyzer™ (Ankom Technology, NY). Particle size was determined using the Penn State particle separator and the procedure of Heinrichs and Kononoff (2002). The pH of DDGS and barley were determined using the procedure of Jasaitis (1987).

Statistical Analysis

Rumen fermentation parameters (pH, VFA concentration, osmolality, ammonia) were analyzed using the PROC MIXED procedure of SAS 9.1 (SAS Institute Inc. 2003), accounting for repeated measures with the random effect of heifer and the fixed effect of treatment. Polynomial orthogonal contrasts were used to determine significant linear, quadratic and cubic effects. The Kenward Roger adjustment on denominator degrees of freedom was used and significance declared at $P < 0.05$. Trends are discussed for $P < 0.10$. Regression analysis was carried out on significant contrasts to determine equations for linear, quadratic and cubic effects of DDGS inclusion rates. Furthermore equations were used to determine theoretical minima and maxima for each significant parameter.

Results and Discussion:

While not significant different, the highest mean ruminal pH averaged over the course of a 24 h period was associated with the rolled barley treatment while the lowest was associated with the 25%DDGS pellet (Table 5.2). Failure to see a treatment effect was

unexpected as the substitution of a non-starch concentrate for a starch-based concentrate should theoretically result in reduced microbial fermentation activity, altered VFA production pattern including lower overall levels and increased rumen pH (Ørskov, 1986).

As in trial 2, we examined not only mean rumen pH as an indicator of rumen acidosis, but also evaluated the length of time over a 24 h period that rumen pH is below a critical value. Critical pH values used where time below 5.8 (mild), 5.6 (moderate) and below 5.2 (severe; acute; Nocek, 1997; Krause *et al.* 2002; Penner *et al.* 2007). Table 5.2 gives the results of the data analyzed in this fashion. Consistent results are observed across all 3 pH cutoff points in that mean rumen pH was lower for pelleted barley vs. rolled barley and that addition of 15 or 25% to pelleted barley also depressed mean rumen ph at all 3 cutoff points relative to rolled barley. In contrast addition of 35% DDGS to pelleted barley elevated rumen pH to values that were not different from that of rolled barley.

These results mirror that of Trial 2 where substitution of DDGS for rolled barley at intermediate levels (i.e. 7 & 14 % DM) failed to moderate pH decline, while substitution of 21% DDGS for rolled barley brought pH back to levels associated with the rolled barley diet. Pelleted barley is known to be rapidly fermented and can stress rumen acid-base metabolism to a much greater degree than rolled barley (McKinnon & Williams unpublished). The greater stress on rumen metabolism is evident from the present work in that mean rumen pH for rolled vs. pelleted barley was 6.1 vs. 5.63. In trial 2 it was hypothesized that the negative effect of DDGS on rumen pH was attributed to specific characteristics of wheat-based DDGS including the fact that it is an acidic feedstuff (initial pH ~4.x) with a very small particle size. Thus although it is a high fibre feed, its effectiveness at stimulating chewing activity and saliva secretion and thus rumen buffering capacity is limited. In trial 2, addition of 21% DDGS to rolled barley led to rumen fermentation conditions similar to that of the rolled barley treatment. In this trial, rumen pH was improved at the 35% level of DDGS. The fact that it took more DDGS to improve rumen pH in this trial is likely attributed to the greater stress placed on rumen pH due to the pelleted nature of the barley.

Fermentation Characteristics (VFA, Ammonia and Osmolality)

Table 5.3 gives the VFA and ammonia N concentrations of the rumen fluid from the heifers fed the 5 dietary treatments. Total VFA and ammonia N levels showed typical diurnal variation, increasing after feeding and declining with increasing time post-feeding. These patterns are consistent with feeding barley-grain based diets and have been shown by other researchers (Krause *et al.* 1998). Ruminal NH_3 N values averaged were low for the rolled and pelleted barley treatments. Rumen ammonia levels increased with all 3 addition rates of DDGS to the pelleted barley treatment. This is a reflection of the higher CP content of these treatments.

Table 5.1. Diet composition and analysis for rolled vs. pelleted barley/DDGS treatments

	Treatment				
		Pelleted Barley			
	Rolled Barley	0% DDGS	15% DDGS	25% DDGS	35% DDGS
Diet Composition (% DM basis)					
Silage	6.2	6.2	6.2	6.2	6.2
Supplement	5.5	5.5	5.5	5.5	5.5
Rolled Barley	88.4	-	-	-	-
Pelleted Barley	-	88.4	73.4	63.4	53.4
Wheat DDGS	0.0	0.00	15.0	25.0	35.0
Supplement Composition (% DM basis)					
Canola Oil	3.5	3.5	3.5	3.5	3.5
Canola Meal	50.7	50.7	50.7	50.7	50.7
Limestone	22.9	22.9	22.9	22.9	22.9
LS106[Z]	8.6	8.6	8.6	8.6	8.6
Rumensin Premix[Y]	7.2	7.2	7.2	7.2	7.2
Trace mineral salt[X]	7.1	7.1	7.1	7.1	7.1
Ration Analysis (% DM basis)					
CP	10.8	10.9	15.0	17.9	21.1
ADF	7.3	6.7	8.6	10.0	11.5
NDF	18.7	15.3	16.6	17.8	20.8
Starch	60.9	64.2	52.8	48.1	39.2

[Z] LS 106 44 500 IU vitamin A, and 88 000 IU vitamin D_3 kg^{-1}; [Y] Rumensin premix: 3% monensin sodium.

[X] Trace mineral salt: 95% sodium chloride, 12 000 ppm zinc, 10 000 ppm manganese, 4 000 ppm copper, 400 ppm iodine, 60 ppm cobalt, 30 ppm added selenium.

Table 5.2 Dry matter intake and rumen pH measurements of cattle fed rolled vs. pelleted barley / DDGS combinations.

	Treatment						
		Pelleted Barley					
	Rolled Barley	0% DDGS	15% DDGS	25% DDGS	35% DDGS	SEM	$P_{Treatment}$
Dry Matter Intake kg d^{-1}	7.77	7.28	7.37	7.25	7.68	0.340	0.75
Mean Daily Rumen pH							
In-dwelling pH	6.10	5.63	5.53	5.39	5.99	0.241	0.18
Rumen pH Parameter 5.8 or lower							
Mean pH	5.34a	5.00b	5.04b	5.00b	5.24ab	0.064	0.002
Total time (min)	499	926	1055	980	625	180.0	0.15
Time between 5.5 and 5.8 (min)	298	34	387	331	446	138.2	0.31
Rumen pH Parameter 5.5 or lower							
Mean pH	5.24a	4.95c	4.99bc	5.01bc	5.17ab	0.050	0.003
Time between 5.2 and 5.5 (min)	95	339	250	308	67	91.1	0.17
Rumen pH Parameter 5.2 or lower							
Mean pH	5.06a	4.89b	4.91ab	4.93ab	5.05a	0.042	0.01
Time below 5.2 (min)	107	553	418	340	112	128.0	0.10

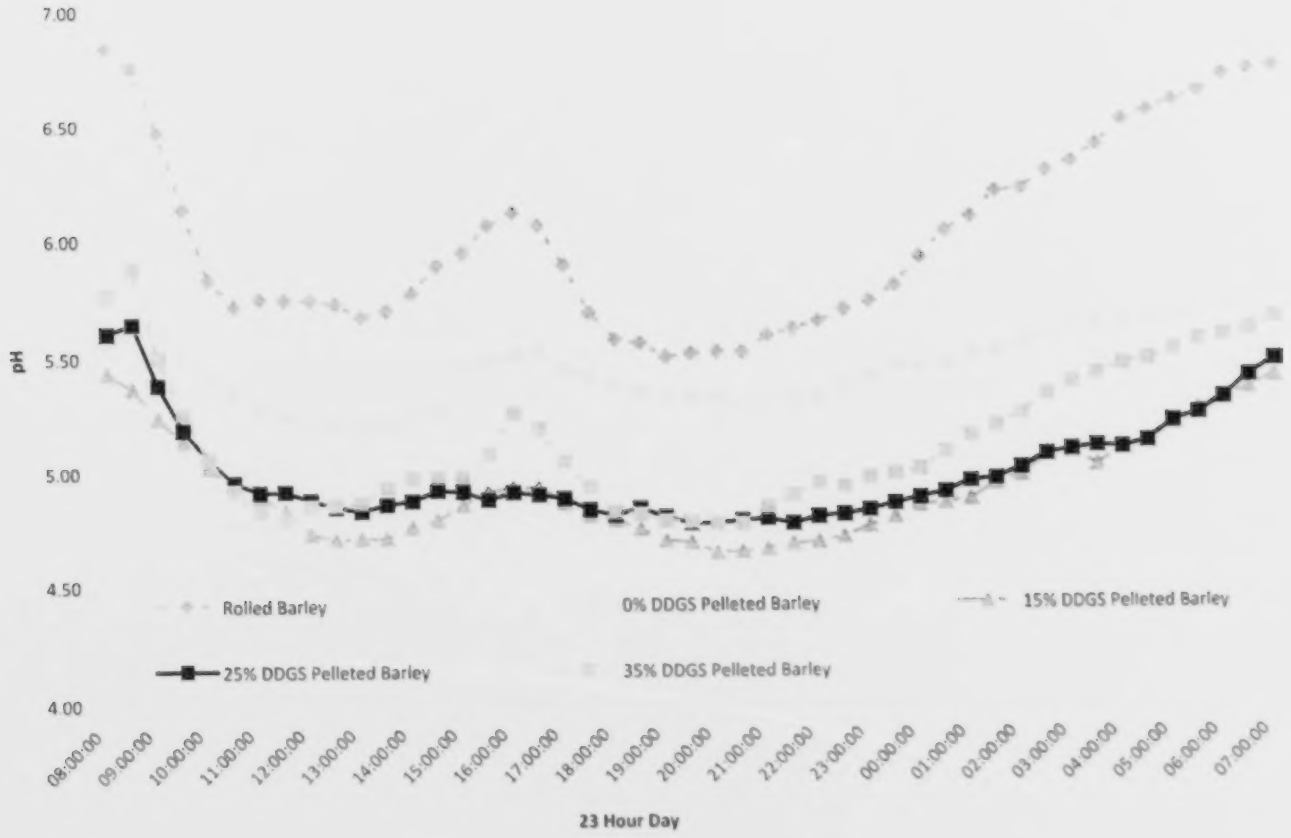

Figure 5.1 Effects of pelleted barley/DDGs on rumen ph.

Table 5.3 Effects of feeding rolled vs. pelleted barley / DDGS combinations on rumen VFA and ammonia N levels.

	Treatment							
		Pelleted Barley						
	Rolled Barley	0% DDGS	15% DDGS	25% DDGS	35% DDGS	*SEM*	$P_{treatment}$	P_{time}
Total VFA (mmol)	88.15	105.97	92.25	88.18	95.94	11.610	0.8	0.03
Acetate (mmol)	39.38	43.91	38.90	37.56	42.34	4.771	0.87	0.22
Propionate (mmol)	32.46	39.79	36.70	33.17	35.95	5.342	0.87	0.54
A:P Ratio	1.29	1.32	1.16	1.24	1.28	0.130	0.91	0.08
Butyrate (mmol)	10.72	15.13	10.85	11.54	11.31	1.770	0.4	0.0001
IsoButyrate (mmol)	0.80	0.71	0.67	0.64	0.72	0.070	0.62	0.04
Valerate (mmol)	3.04	3.58	3.09	3.43	3.57	0.550	0.92	0.06
IsoValerate (mmol)	1.30	0.90	0.93	0.91	1.07	0.110	0.08	0.42
Spot Sample pH	5.97	5.32	5.59	5.48	5.66	0.152	0.07	0.0001
Ammonia (mg dL^{-1})	2.51b	1.72b	4.16a	5.24a	4.92a	0.626	0.004	0.0001

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